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COMPARATIVE ANALYSIS OF BIOLOGICALLY ACTIVE SUBSTANCES OF *VERBASCUM PHLOMOIDES AND HYPERICUM PERFORATUM* L. IN DIFFERENT ECOSYSTEMS

Abstract. A comparative analysis of the biologically active substances content in plant raw materials *Verbascum phlomoides* and *Hypericum perforatum* depending on their habitat within the Stryi district (anthropogenic, plain, foothill and mountain ecosystems) was conducted. The content of ascorbic acid, chlorophylls a and b, carotenoids in plant raw materials *Verbascum phlomoides* and *Hypericum perforatum* was determined spectrophotometrically. The highest content of ascorbic acid was found in the raw materials of *Verbascum phlomoides* and *Hypericum perforatum* in the plain ecosystem. The concentration and content of chlorophylls and carotenoids in the studied plant raw material were studied, their highest content in plant raw materials *Verbascum phlomoides* in the plain and mountain ecosystems, and in *Hypericum perforatum* – in the plain ecosystem was found. The research results show that the in the anthropogenically loaded habitat of plants leads to a decrease in the content of ascorbic acid and photosynthetic pigments in both studied plants. This indicates a significant anthropogenic impact on the environment. Comparative analysis shows that the most suitable ecosystems for harvesting wild medicinal raw materials *Verbascum phlomoides* and *Hypericum perforatum* are the plains of Stryj district.

Key words: biologically active substances, raw materials, *Verbascum phlomoides, Hypericum perforatum*, ecosystem, ascorbic acid, carotenoids.

Harvesting of plant material is possible both from the wild flora and introduced into the culture. Many medicinal plants are used for veterinary and medical purposes. These raw materials are harvested directly by farm experts. It should be remembered that there is no endless supply of plants in nature, which means that the harvesting of raw materials requires a considered and rational approach to this issue. Considered approach to the harvesting of wild plants. It is necessary to preserve their fertility, yields, stocks, and, if possible, expand the areas of their growth. Based on this, the procurement of wild medicinal raw materials should be considered and properly considered. The studies on the plants have been conducted for a long time and now we have all the data on the qualitative and quantitative composition of a particular plant and plant development [2; 4; 14].

Therefore, *Hypericum perforatum* is a medicinal plant with a high content of secondary metabolites. It contains such biologically active substances as xanthones, flavonoids, petrodiatrons, phenolic compounds. *Hypericum perforatum* has antiviral, antioxidant, antidepressant, antimicrobial and anti-inflammatory effects [11]. The concentration of biologically active components in plants varies depending on their habitat and environmental factors. *H. perforatum is* usually not cultivated, it is harvested in the wild. It plays a significant role in the difference between herbal and synthetic drugs. Plants should be harvested in the open air. It is also necessary to collect plants during the flowering period, because then the maximum amount of nutrients is concentrated in them [7] *Verbascum phlomoides* is a medicinal plant that contains such biologi-

cal substances as flavonoids, saponins, glycosides. Iridoid glycosides. Extracts from *Verbascum phlomoides* are used for diseases of the upper respiratory tract, for colds on coug [17]. *Verbascum phlomoides* is used in herbal medicine, it has anti-inflammatory and antimicrobial effects. Medications from *Verbascum phlomoides* have the ability to reduce tissue swelling and convulsions. They are used in asthma, pneumonia, and hemoptysis.

Bioactive substances are of particular importance in the development of products in the pharmaceutical, medical, chemical and food industries. Scientists conduct studies with biologically active substances to test their effects on humans and participate in biochemical reactions [5; 15]. Interest in the production of bioactive compounds from natural resources is increasing. Natural raw materials play an important role in the search for new pharmacologically active compounds. For centuries, medicinal plant products have proven to be one of the main natural sources with therapeutic properties. Currently, the detection of bioactive substances in plants is a lengthy and painstaking process. It includes chemical, biological and molecular methods that provide important results in the prevention and control of various diseases [1; 17].

MATERIALS AND METHODS

The Stryi region is characterized by a temperate continental climate. It is a forest- steppe area dominated by plains, but there are foothills and mountainous parts. The Stryi region is characterized by low soil fertility. Soils are acidic with low oxygen level. There are oil and gas fields, water intakes and mining outlets in the area. The natural resources of which the Stryi region is rich are oil and gas, mineral and underground water, rich beech forests.

The studies were carried out with samples: *Verbascum phlomoides*, *Hypericum perforatum* sampled in different ecosystems in the Stryi region. The plant material was collected in five ecosystems: anthropogenically loaded (near Morshyn), plain (near Pidhirtsi village), foothills (near Verkhnya Lukavytsya), mountainous (near Nizhnyaya Stinawa) and lowland (outskirts of Dovholuka village). Medical raw materials were collected in the summer-autumn period 2019 – 2021. Raw materials were collected in wild populations. Flowers were used for the study. Accordingly, the flowers were collected at the end of flowering (July-August). Several individuals of each species of the same age group were selected in satisfactory sanitary condition in accordance with the methods. Samples were taken in clean areas, far from roads. When sampling, all the rules of proper sampling of medicinal raw materials were observed: plants were harvested in dry weather between 12:00 and 17:00; raw materials were sampled in paper bags and on- site labeled, the date, sample number, time and place of collection were written. The selected samples were packed in plastic bags, signed and placed in the freezer for storage.

In vegetable raw materials *Verbascum phlomoides* and *Hypericum perforatum* the content of vitamin C, the content of photosynthetic pigments was determined in plant homogenates (photometer UNICO 2150). The studies were conducted on the Polonia Academy in Czestochowa.

RESULTS AND DISCUSSION

Analysis of the ascorbic acid content in plant raw materials. Many medicinal plants are depots of important biologically active substances, so they are often used in medical practice, and their biologically active substances determine the effectiveness of treatment. These include ascorbic acid, which is an indicator of the directivity of redox processes and has antioxidant activity, determining the resistance of plants.

Ascorbic acid affects the growth and development of cells. It also promotes better absorption of calcium by the body. The body consumes a large amount of ascorbic acid in the fight against disease and in the healing of wounds [3; 10]. The human body cannot accumulate or synthesize vitamin C, so it must be included in the daily diet for normal functioning of the body [11].

In the scientific field, there are continuing disputes over improving dosage rates of ascorbic acid to maintain its level in the human body. Today, scientists worry that fruits and vegetables are not an environmentally friendly and sufficient source of the right dose of ascorbic acid. Since ascorbic acid is not synthesized in humans, but is indispensable in all biochemical processes, it can be obtained from plants [9]. Plants are considered reliable on a quality source of vitamins. The fruits of rose hips, black currant, sea buckthorn, pine needles, and green walnut fruits are rich in ascorbic acid. The search for new sources of naturally occurring biologically active substances is currently underway, so the first stage of the study was to analyze the content of ascorbic acid in the plant material of *Hypericum Perforatum* and *Verbascum Phlomoides*. The results of determining the content of ascorbic acid are presented in table 1.

Table 4. The content of ascorbic acid in vegetable raw materials *Hypericum Perforatum and Verbascum Phlomoides*, AK, mkg/l (M ± m)

№	The name of the ecosystem	Hypericum Perforatum	Verbascum Phlomoides
1.	Anthropogenically loaded (outskirts of Morshyn city)	4.53±0.465	1.58±0.213
2.	Foothills ecosystem (outskirts of Verkhny Lukavitsa village)	4.97±0.414	2.97±0.247
3.	Flat ecosystem (outskirts of Pidhirtsi village)	6.04±0.503	3.84±0.32
4.	Mountain ecosystem (outskirts of village of Nyzhnia Stinava)	5.58±0.377	2.56±0.131
5.	Lowland ecosystem (outskirts of Dovholuk village)	5.89±0.490	3.05±0.254

The table above shows that the content of ascorbic acid in the flower extracts of *Verbascum Phlomoides* ranges from 1.58 ± 0.213 to 3.84 ± 0.32 μg/l (p0.01-0.05). Whereas the content of ascorbic acid in the extracts of flowers of *Hypericum Perforatum* ranges from 4.53 ± 0.465 to 6.04 ± 0.503 mkg/l (p0.01-0.05).

The content of ascorbic acid in the extracts of flowers *Verbascum Phlomoides* ranges from 1.58 ± 0.213 to 3.84 ± 0.32 μg/l (p0.01-0.05). It was found that the lowest content of AA in anthropogenically loaded terrain (the outskirts of Morshyn city).

According to the studies of Voloshin A. A. [16] the quantitative composition of ascorbic acid in the studied samples of divinity by the titrimetric method in the flowers was 2.25 ± 0.10 mkg/l (p<0.01-0.05). Therefore, we can conclude that the samples of *Verbascum Phlomoides* we study are more suitable for use in the pharmaceutical field, since the amount of ascorbic acid in them varies within 3.84 ± 0.03 mkg/l (p<0.01-0.05).

Studies have shown that the lowest content of ascorbic acid in raw materials *Hypericum perforatum* is found in the anthropogenically loaded ecosystem (Morshin vicinity), which is 4.53 ± 0.465 μg/l (p0.05). and is 6.04 ± 0.503 μg/l (p0.01-0.05).

The lowest vitamin C content in the anthropogenically loaded region is explained by the fact that it is an antioxidant and is consumed to counteract the action of free radicals and to overcome the effects of oxidative stress. The highest level of AK was detected in the plain area (the outskirts of the village of Pidhirtsi), which can be a consequence of favorable climate and soil and climatic conditions.

Therefore, it can be seen from the indicators described above that the content of ascorbic acid in *Hypericum perforatum* is twice as high as that in *Verbascum phlomoides*.

Analysis of chlorophylls and carotenoids in vegetable raw materials. Carotenoids, vitamins C, E, selenium, bioflavonoids (vitamin-like substances contained in the peel of plants) are the most important biological active sybstance. Many plant extracts, vitamins, amino acids, minerals, trace elements exhibit antioxidant properties, or their antioxidant properties, or antioxidants [10].

One of the known biologically active substances in the plant organism are chlorophylls and carotenoids. They are important for determining the analysis of the antioxidant activity of the studied plants. Chlorophyll has the ability to increase the number of red blood cells, because its structure is similar to the structure of hemoglobin. Chlorophyll has the ability to neutralize inflammatory processes in humans. Carotenoids play an important role in protecting the body against disease. They activate the synthesis of cytokines and interleukins of the body's immune system. Carotenoids are capable of slowing the growth and development of malignancies in the living organism. Carotenoids also increase the body's resistance to external factors [10, 12].

The study of the pigment composition of plants is of great importance in the mechanisms of adaptation of plants to environmental conditions. Depending on the light conditions, the number of photosynthetic pigments varies [6]. The results determined the concentrations of chlorophylls a, b i carotenoids were recorded in the figure 1-2.

It is a well-known fact that environmental conditions significantly affect photosynthetic processes in plants, which can affect the concentration of chlorophylls and carotenoids.

The second stage of the study was to investigate the dynamics of changes in the concentration of chlorophylls and carotenoids in the flowers of *Hypericum perforatum* and *Verbascum phlomoides*, depending on growth conditions.

The concentration of chlorophyll a in the tested samples of *Verbascum phlomoides* ranges from 1.43 ± 0.119 to 2.75 ± 0.229 mg/l ($p<0.01$-0.05). The highest concentration of chlorophyll a in the extracts of flowers *Verbascum phlomoides* in the mountain ecosystem is 2.75 ± 0.229mg/l. This may be due to better photosynthesis transpiration processes in the highlands.

The concentration of chlorophyll a in the tested samples of *Hypericum perforatum* ranges from 0.328 ± 0.0273 to 0.713 ± 0.059 mg/l ($p<0.01$-0.05). The highest concentration of chlorophyll a is observed in the flowers of *Hypericum perforatum* in the plain area (the outskirts of the village Pidhirtsi) is 1,975±0.164 mg/l ($p<0.01$-0.05).

The concentration of chlorophylls a and b in plant material in different ecosystems. The concentration of chlorophyll b in the tested samples of *Hypericum perforatum* ranges from 0.985±0.107 to 1,975±0.164 mg/l ($p<0.01$-0.05).

The highest index of chlorophyll b concentration in *Hypericum perforatum* flowers in flat ecosystem is 1,975±0.164 mg/l ($p<0.01$-0.05) and the lowest in the plant material from the lthe anthropogenically loaded ecosystem is 0.985±0.107 mg/l($p<0.01$-0.05). According to the literature, under unfavorable conditions of plant growth, the concentration of chlorophyll b increases. When the concentration of the amount of chlorophyll b changes and the concentration of chlorophyll a.

The concentration of chlorophyll b in the tested samples of *Verbascum phlomoides* ranges from 1,05±0.087 to2,58±0.215mg/l ($p<0.01$-0.05).

The highest index of chlorophyll b concentration in Verbascum phlomoides flowers in mountain ecosystems is 2,58±0.215 mg/l($p<0.01$-0.05), and the lowest in plant material from anthropogenically loaded ecosystems is 1.05±0.087mg/l ($p<0.01$-0.05).

Determination of the content of chlorophylls a and b made it possible to calculate the content of carotenoids in the studied objects and to determine the content of chlorophyll a and b pigments and carotenoids.

The concentration of carotenoid in the tested samples of *Verbascum phlomoides* ranges from 1,48±0.123 to 2,57±0.214 mg/l ($p<0.01$-0.05). The highest carotenoid concentration in *Verbascum phlomoides* flowers in the flat ecosystem (outskirts of Pidhirtsi village) is 2,57±0.214 mg/l ($p<0.01$-0.05), and the lowest in the plant material from the anthropogenically loaded (outskirts of Morshyn city) is 1,48±0.123 mg/l ($p<0.01$-0.05).

The concentration of carotenoid in the tested samples of *Hypericum perforatum* ranges from 2,253±0.187 to 3,834±0.319 mg/l ($p<0.01$-0.05).The highest carotenoid concentration in *Hyperi-*

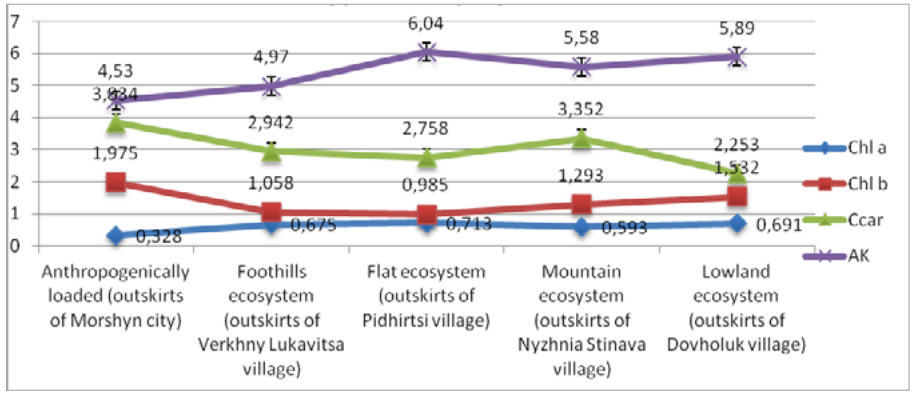

Fig. 1. The concentration of chlorophylls a and b, carotenoids and ascorbic acid in raw materials *Hypericum perforatum*

cum perforatum flowers in the flat ecosystem (outskirts of Pidhirtsi village) is 3,834±0.319 mg/l ($p<0.01$-0.05), and the lowest in plant material from the anthropogenically loaded (outskirts of Morshyn city) is 2,253±0.187 mg/l ($p<0.01$-0.05).

Therefore, carotenoids are the largest of all other photosynthetic pigments studied in vegetable raw *Hypericum perforatum*. The most favorable conditions for growth are found in the mountainous and flat areas. The lowest rates are in the anthropogenically loaded area. This may be due to a large amount of pollution in the area, due to factories and waste emissions and environmental pollution due to the development of the resort in the territory of Morshyn. Chlorophyll a is lower than chlorophyll b. The highest index of chlorophyll a and chlorophyll b is in the lowland area and the lowest in the anthropogenically loaded area. This indicates contamination of the area or adverse climatic conditions. In *Verbascum phlomoides* raw materials, chlorophyll a, b and carotenoid levels fluctuate. The highest in the mountainous and plain areas, and the worst in the anthropogenically loaded area.

The results of the studies indicate a clear decrease in the content of chlorophylls and carotenoids in both studied plants from the outskirts of Morshyn. This indicates significant anthropogenic pressure on the environment. It is not advisable to harvest raw materials in such areas, as

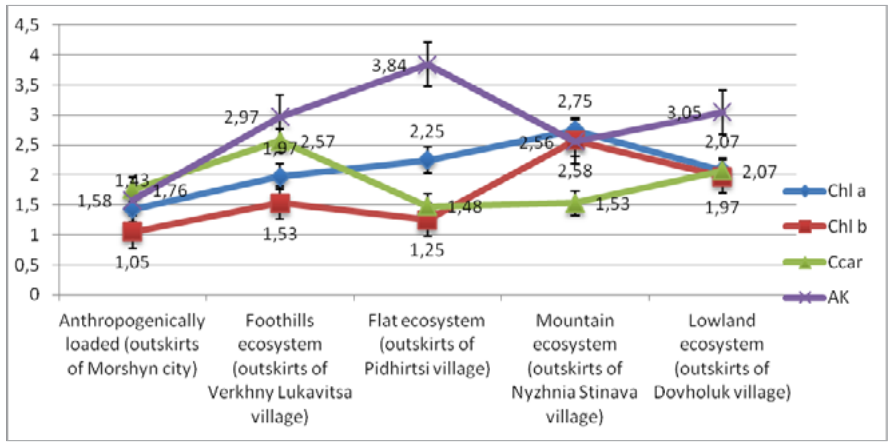

Fig. 2. The concentration of chlorophylls a and b, carotenoids and ascorbic acid in raw materials *Verbascum phlomoides*

heavy metals can also accumulate in plants. By increasing the amount of heavy metals in soil or water, they can cause disruption of physiological processes in plants, especially the process of photosynthesis and the content of photosynthetic pigments [13].

Hypericum perforatum showed high results in the plain ecosystem (outskirts of Pidhirtsi village), mountain ecosystem (outskirts of Nyzhnia Stinava village), lowland ecosystem (outskirts of Dovholuk village). Low rates in *Hypericum perforatum* were in the anthropogenically loaded area.

Verbascum phlomoides had a high ascorbic acid content in the lowland ecosystem, a high carotenoid content was observed in the foothills. All the lowest indicators were observed in the anthropogenically loaded area.

According to studies, it can be concluded that *Hypericum perforatum* performed better than *Verbascum phlomoides*. *Hypericum perforatum* has proven to be more stable to anthropopression and soil conditions. *Hypericum perforatum* showed high performance in almost all the study areas, so it can be said that it is resistant to environmental conditions and climatic conditions. *Verbascum phlomoides* performed well with only a few indicators in the lowland and suburban areas. *Hypericum perforatum* is better collected in flat ecosystem (outskirts of Pidhirtsi village), and *Verbascum phlomoides* is better collected in mountain ecosystem (outskirts of village of Nyzhnia Stinava). Therefore, for further use of these plants as medicinal raw materials for the manufacture of medicines, they can be selected in flat ecosystem (outskirts of Pidhirtsi village) and mountain ecosystem (outskirts of village of Nyzhnia Stinava).

CONCLUSION

The problem of adaptation of plants to changes in the environment is becoming more urgent. Plants cannot avoid change, because they have an attached way of life. Now with increasing anthropogenic pressure and global changes in climate, plants are more difficult to adapt than ever. Therefore, high stress resistance and antioxidant activity is as important to the plant organism as ever. The issues of adaptation of plant natural communities and the knowledge of the mechanism of their adaptation are mainly related to the conservation and restoration of biodiversity.

As a result of determining the content of biologically active substances in the investigated plants within the Stryi district of *Verbascum phlomoides* and *Hypericum perforatum*, the following conclusions are reached. Indicators of ascorbic acid content, photosynthetic pigments and catalase activity were determined. The plants were selected from 5 different ecosystems in the Stryi region. and ascorbic acid content was found to be twice as high in *Hypericum Perforatum* as in *Verbascum Phlomoides*. The highest rates of ascorbic acid were in the outskirts of the village of Pidhirtsi. The high content of ascorbic acid indicates the high value of vegetable raw materials and therefore the high antioxidant activity.

The concentration of photosynthetic pigments plays an important role in protecting plants from environmental conditions. High levels of chlorophylls and carotenoids were found in the mountain ecosystem of the lower Stinava village. Higher scores were found in *Verbascum phlomoides*.

The content of photosynthetic pigments in the investigated plants was determined. The highest content of chlorophyll a, b and carotenoids in the studied raw material of *Verbascum phlomoides* in the lowland and mountain ecosystems. The highest content of chlorophyll a, b and carotenoids in the studied raw material of *Hypericum perforatum* in the lowland ecosystem. Studies show a decrease in the content of chlorophylls and carotenoids in both plants under study in the anthropogenically loaded area. This indicates a significant anthropogenic impact of ha environment. It is not recommended to collect medicinal raw materials in such areas as the plants may be contaminated with heavy metals. Heavy metals disrupt the physiological functions of plants, can pass into medicines and cause harm to health.

As a result of the research, it was found that the best territory for the selection of medicinal plants is the flat area around the village of Pidhirtsi. *Verbascum phlomoides* were found to be better compared to *Hypericum perforatum*. Therefore, *V. phlomoides* selected from the plain ecosystem of the Pidgirtsi village of Stryi district is more promising for use in pharmacological and other fields.

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АНОТАЦІЯ

ПОРІВНЯЛЬНИЙ АНАЛІЗ ВМІСТУ БІОЛОГІЧНО АКТИВНИХ РЕЧОВИН *VERBASCUM PHLOMOIDES I HYPERICUM PERFORATUM* L. ЗАЛЕЖНО ВІД МІСЦЕЗРОСТАННЯ У РІЗНИХ ЕКОСИСТЕМАХ

Пошук екологічно чистих екосистем для заготівлі дикорослої лікарської рослинної сировини є актуальною проблемою фармації. Проведено порівняльний аналіз вмісту біологічно активних речовин у рослинній сировині Verbascum phlomoides та Hypericum perforatum залежно від місця зростання у межах Стрийського району (антропогенна, рівнинна, передгірська та гірська екосистеми). Спектрофотометрично визначено вміст аскорбінової кислоти, хлорофілів a і b, каротиноїдів у рослинній сировині Verbascum phlomoides та Hypericum perforatum та оцінено стан антиоксидантного захисту за активністю антиоксидантних ензимів.

Найвищий вміст аскорбінової кислоти встановлено у сировині Verbascum phlomoides та Hypericum perforatum у рівнинній екосистемі. Досліджено концентрацію та вміст хлорофілів і каротиноїдів у досліджуваній рослинній сировині, встановлено їх найвищий вміст у рослинній сировині Verbascum phlomoides у рівнинній та гірській екосистемах, а у Hypericum perforatum – у рівнинній екосистемі. Результати досліджень свідчать, що місцезростання рослин в антропогенно навантаженій зоні призводить до зниження вмісту аскорбінової кислоти та фотосинтезуючих пігментів в обох дослідних рослинах. Це свідчить про значний антропогенний вплив на навколишнє середовище. Проведені дослідження стану антиоксидантного захисту рослин вказують на вищу стресостійкість Verbascum phlomoides до негативних факторів навколишнього середовища. Порівняльний аналіз свідчить, що найкращими екосистемами для заготівлі дикорослої лікарської сировини Verbascum phlomoides та Hypericum perforatum є рівнинна територія Стрийського району. Перспективною для використання у фармакології є Verbascum phlomoides.

Ключові слова: біологічно активні речовини, лікарська сировина, Verbascum phlomoides, Hypericum perforatum, екосистема, аскорбінова кислота, каротиноїди.